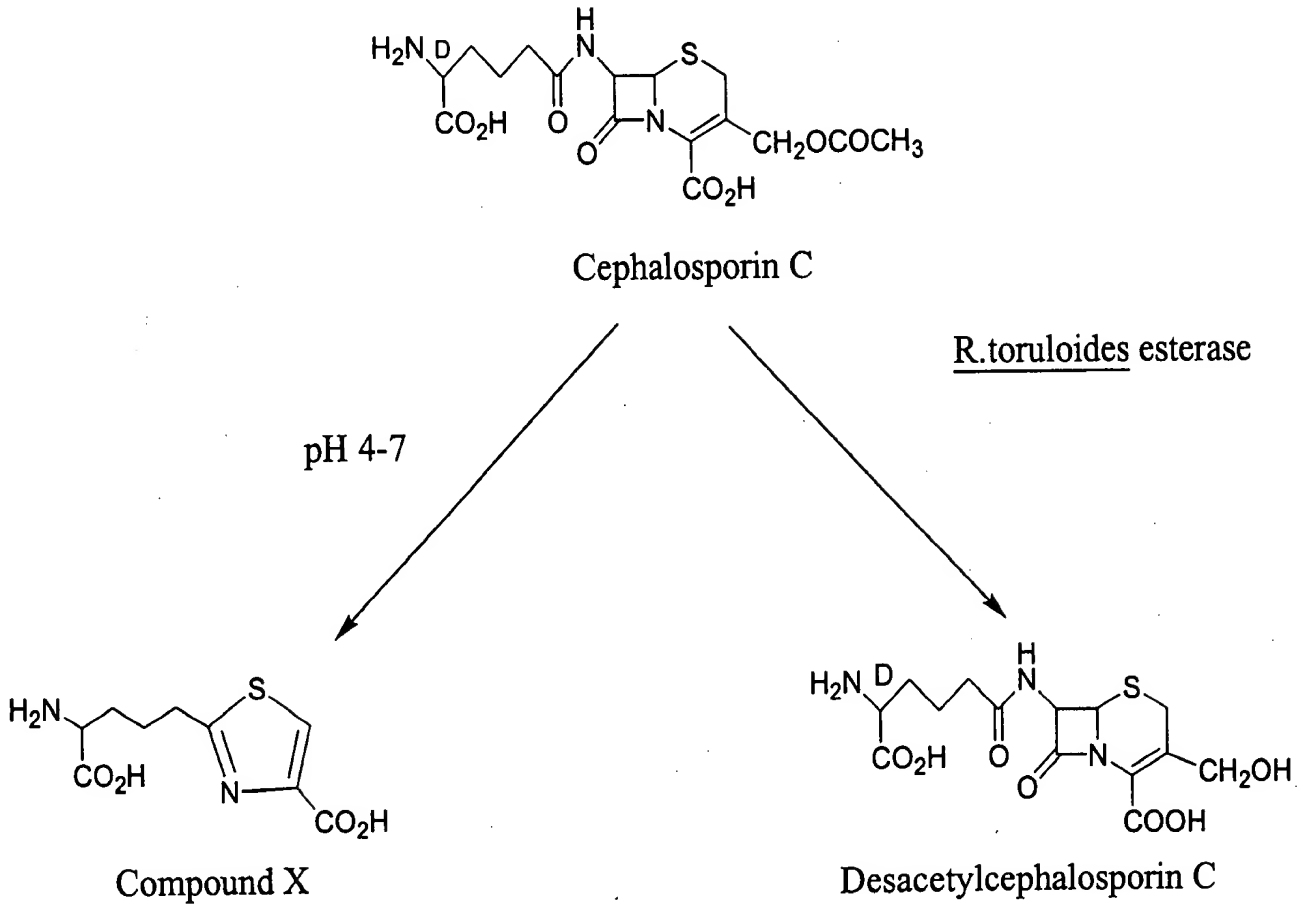


FIG. 1

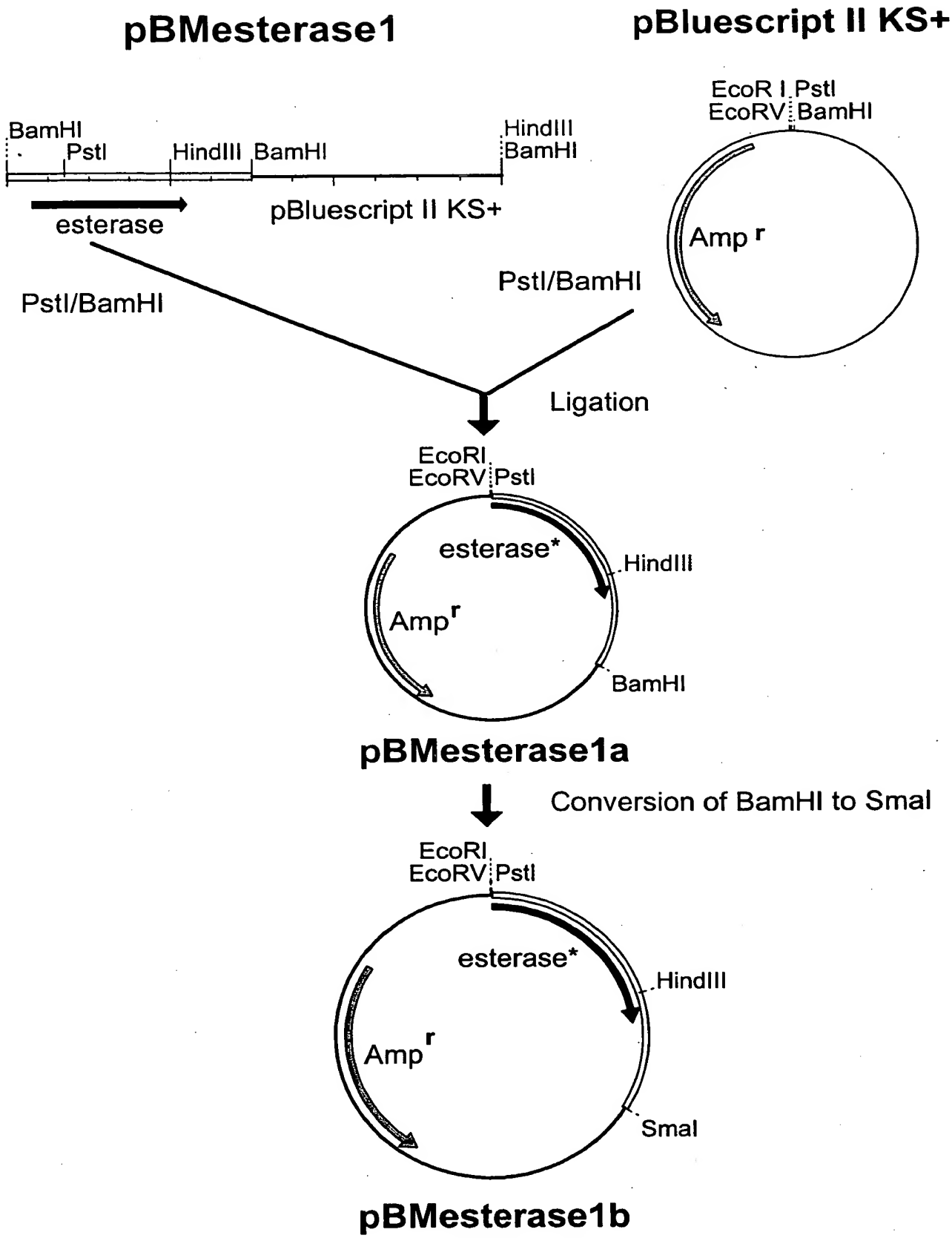
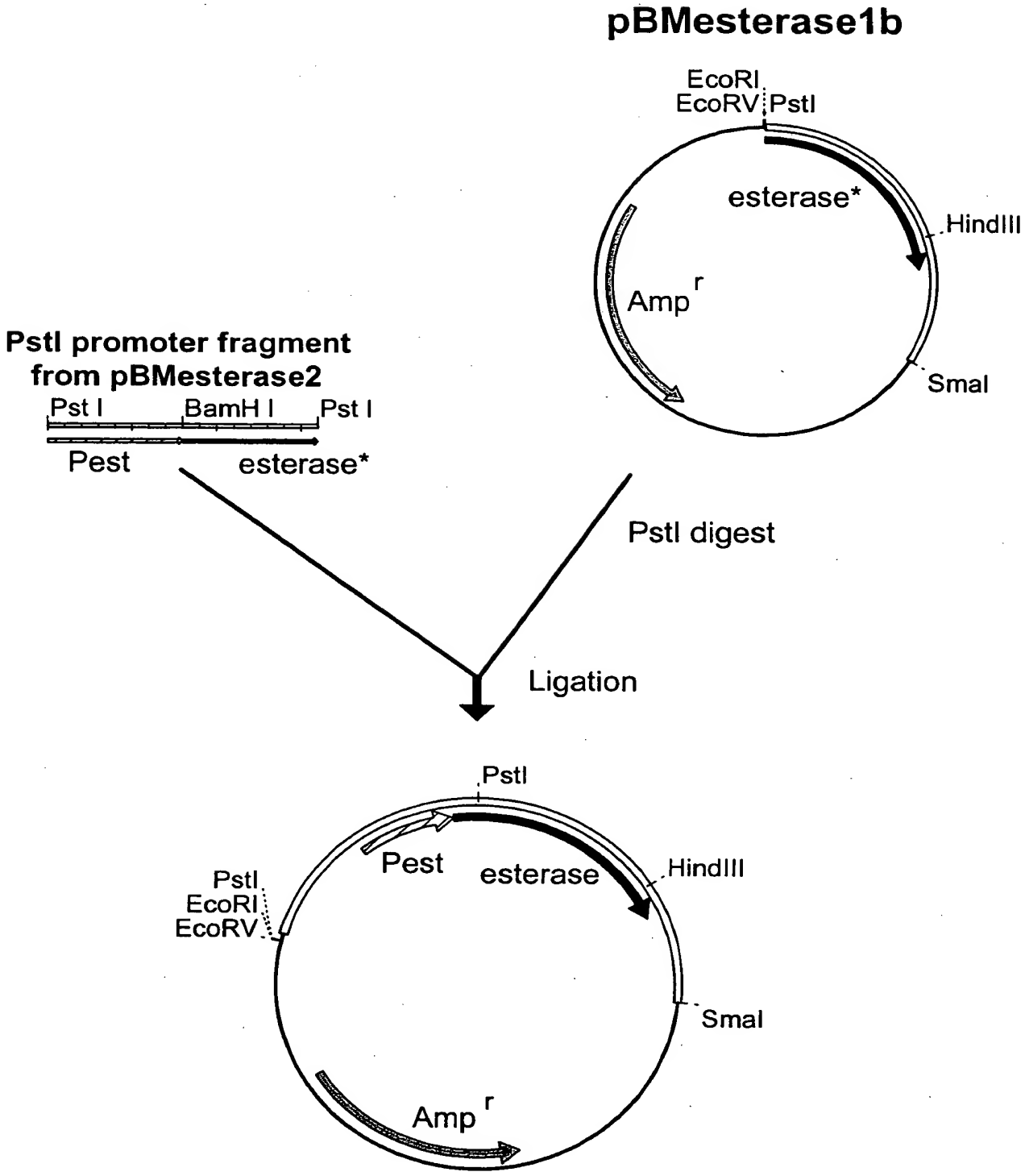


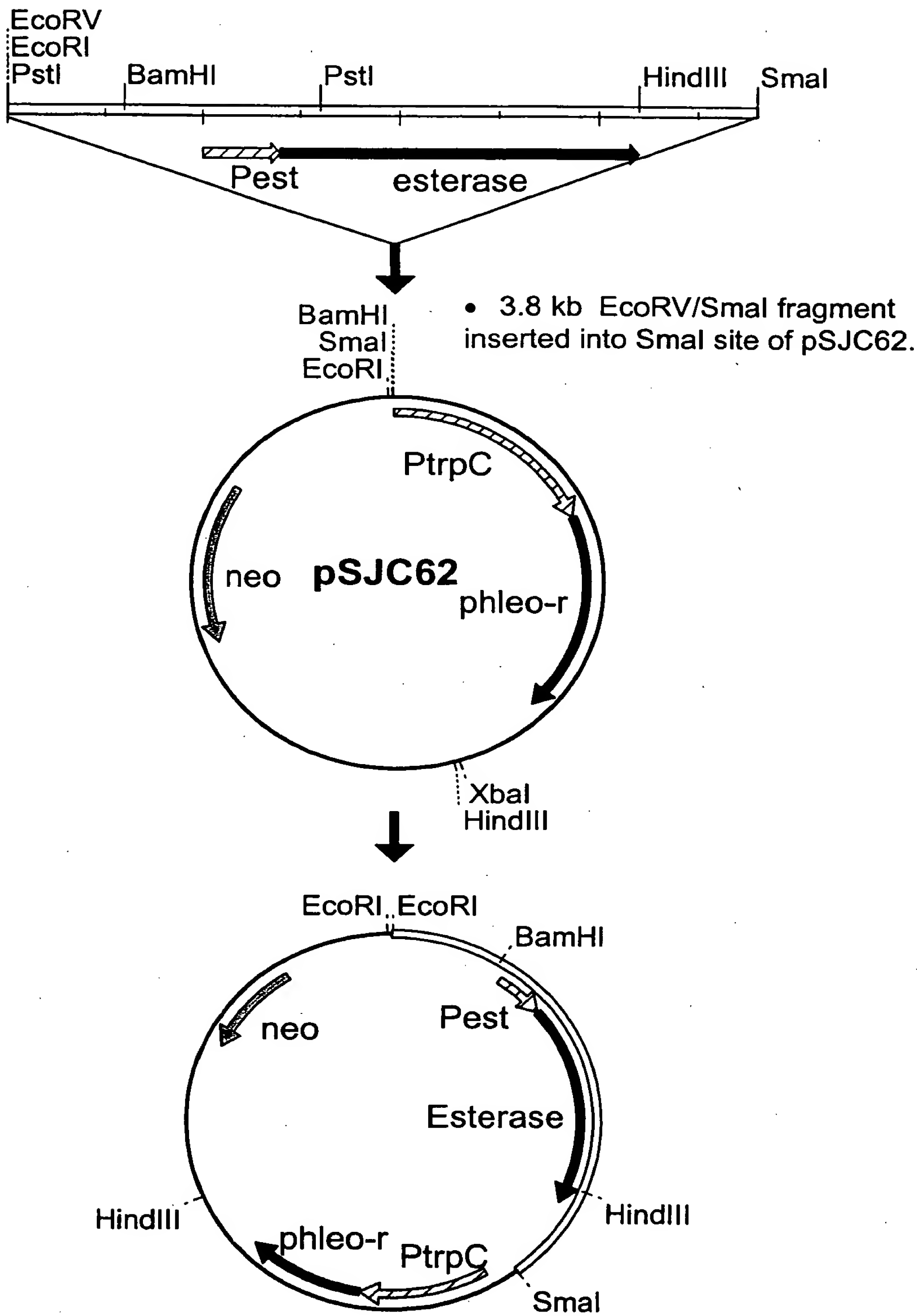
FIG. 2



pBMesterase3

FIG. 3

SmaI/ EcoRV esterase fragment from pBMesterase3



pSJC62.3

FIG. 4

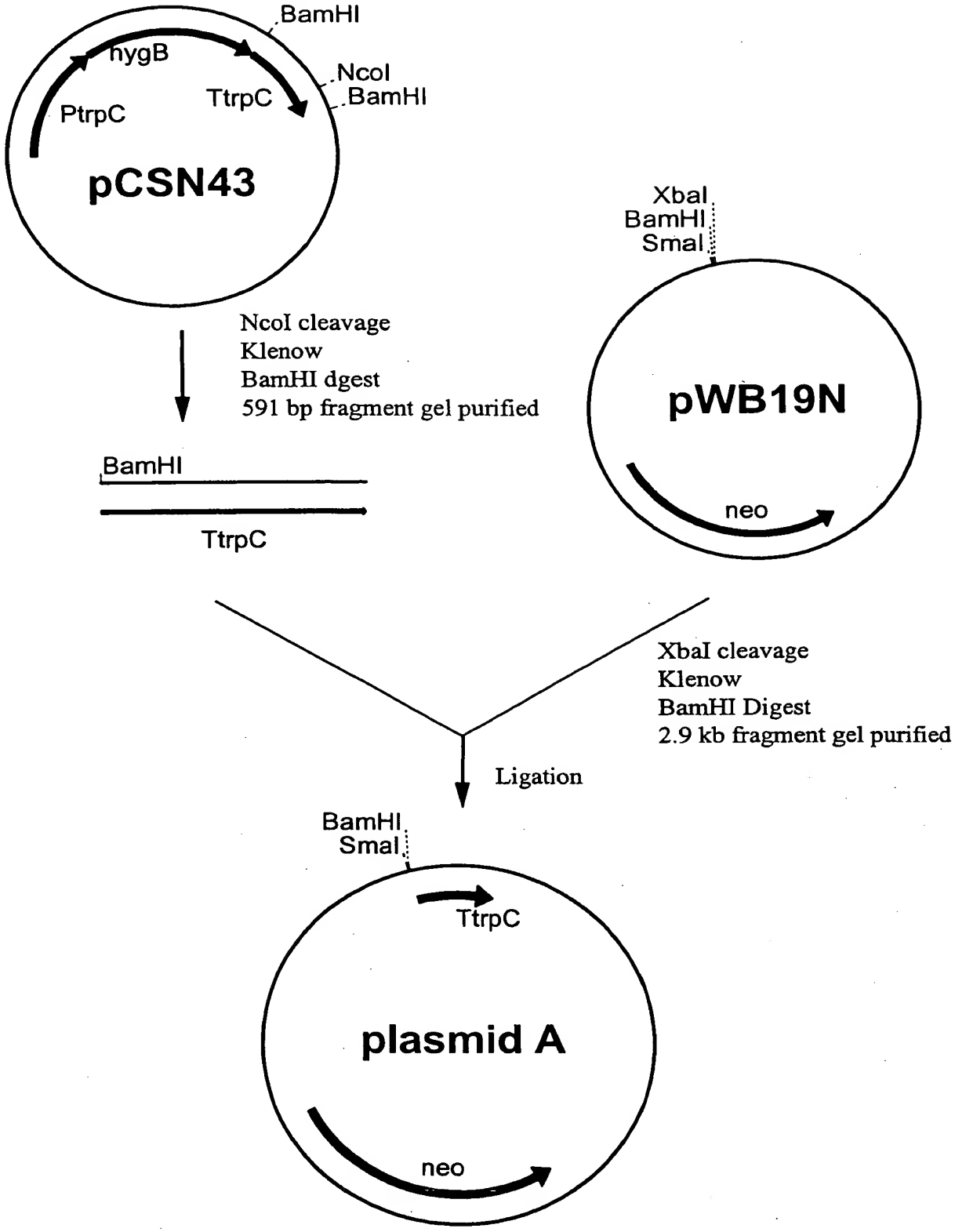
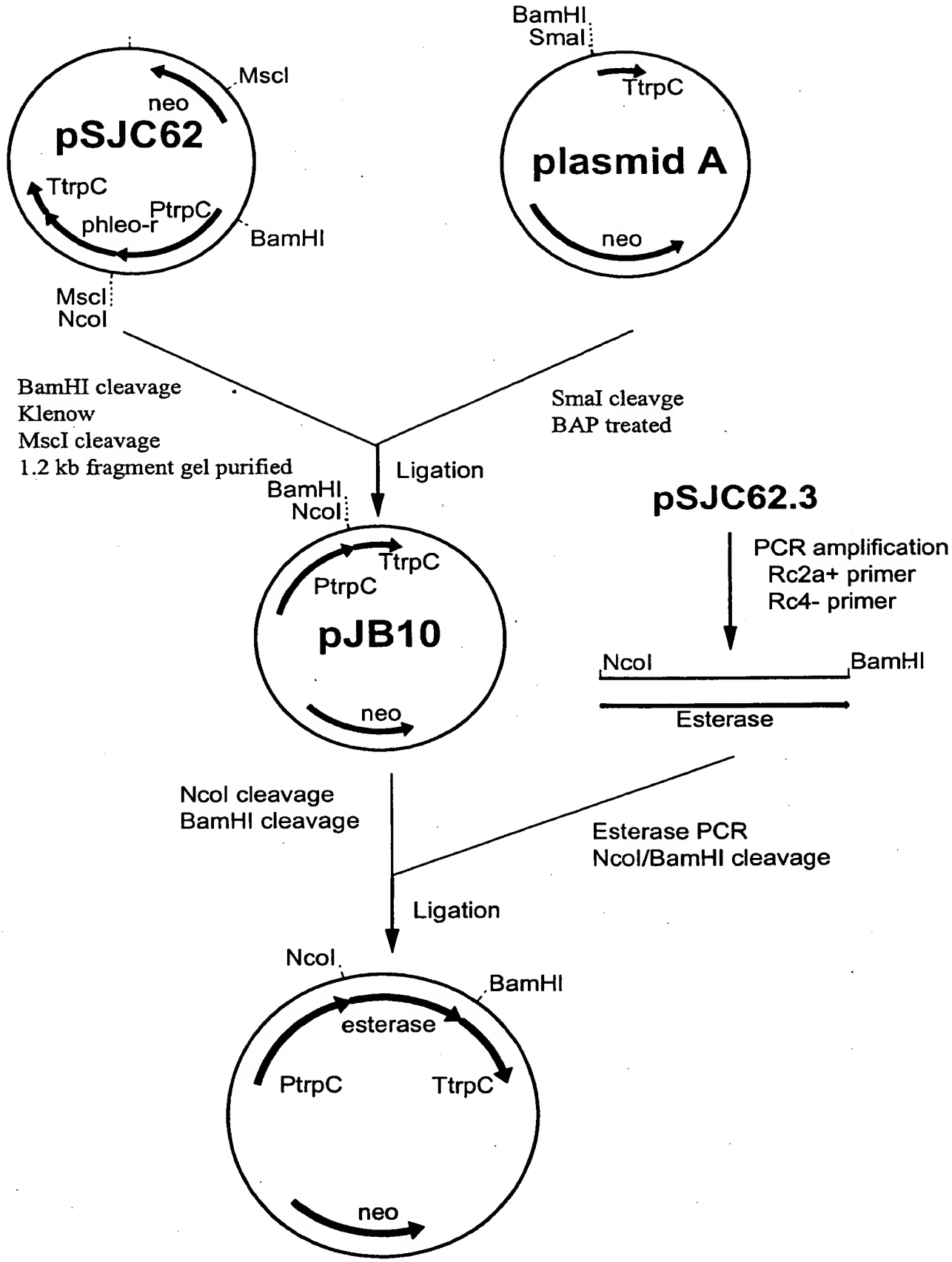


FIG. 5



pBMesterase11

FIG. 6

N-TERMINAL AMINO ACID SEQUENCE

AMINO ACID SEQ.		**T N P N E P**
REV. TRANSLATION		**ACX AAPy CCX AAPy GAPu CC**
INVERSE		**GGPy TCPu TTX GGPu TTX GT**
PROBE	**1**	**GGPy TCPu TTG GGPu TTX GT**
	2	**A**
	3	**T**
	4	**C**

Four 17-mer oligonucleotide probes each with a 32-fold degeneracy were synthesized from the N-terminal amino acid sequence and used to probe a Southern blot of *R.toruloides* DNA.

FIG. 7

RHODOSPORIDIUM ESTERASE cDNA

```
ATGCTCCTTAACCTCTTCACCCTCGCCTCCCTCGCTGCGACGCTCCAGCTCGCCTTTGCC      70
M  L  L  N  L  F  T  L  A  S  L  A  A  T  L  Q  L  A  F  A

TCTCCGACCTCCCTCGTCCGCCGCACGAACCCAAACGAGCCCCCTCCCGTCGTCGACCTC     130
S  P  T  S  L  V  R  R  T  N  P  N  E  P  P  P  V  V  D  L

GGCTACGCCCGCTACCAAGGCTACTTGAACGAGACCGCCGGACTCTACTGGTGGCGCGGA     190
G  Y  A  R  Y  Q  G  Y  L  N  E  T  A  G  L  Y  W  W  R  G

ATCCGCTACGCCTCGGCTCAGCGCTTCCAGGCTCCTCAGACGCCCGCGACGCACAAGGCC     250
I  R  Y  A  S  A  Q  R  F  Q  A  P  Q  T  P  A  T  H  K  A

GTCCGCAACGCGACTGAGTATGGACCGATCTGTTGGCCGGCTAGCGAGGGAACCAACACG     310
V  R  N  A  T  E  Y  G  P  I  C  W  P  A  S  E  G  T  N  T

ACCAAGGGCTTGCCGCCGCCTAGCAACAGCTCGAGCAGCGCGCCGCAGAAACAGGCGTCG     370
T  K  G  L  P  P  P  S  N  S  S  S  S  A  P  Q  K  Q  A  S

GAGGATTGCCTCTTCCTCAATGTCGTTGCCCCCGCCGGCTCGTGCGAGGGCGACAATCTT     430
E  D  C  L  F  L  N  V  V  A  P  A  G  S  C  E  G  D  N  L

CCCGTCCTCGTCTACATTCACGGAGGTGGCTACGCCTTCGGCGATGCGAGCACCGGCAGC     490
P  V  L  V  Y  I  H  G  G  G  Y  A  F  G  D  A  S  T  G  S

GACTTTGCCGCCTTCACCAAGCACACGGGAACCAAGATGGTCGTTGTAAATCTCCAGTAC     550
D  F  A  A  F  T  K  H  T  G  T  K  M  V  V  V  N  L  Q  Y

CGTCTCGGCAGCTTTGGTTTCCTCGCTGGCCAAGCCATGAAGGACTACGGTGTAACGAAC     610
R  L  G  S  F  G  F  L  A  G  Q  A  M  K  D  Y  G  V  T  N

GCCGGCTTGCTTGACCAGCAATTCGCCCTTCAATGGGTTCAACAGCACGTCTCGAAGTTC     670
A  G  L  L  D  Q  Q  F  A  L  Q  W  V  Q  Q  H  V  S  K  F

GGCGGCAACCCCGATCACGTTACGATTTGGGGCGAGTCTGCAGGCGCAGGGTCCGTTATG     730
G  G  N  P  D  H  V  T  I  W  G  E  S  A  G  A  G  S  V  M

AACCAGATCATTGCGAACGGCGGCAACACCGTCAAGGCTCTCGGTCTCAAGAAGCCCCTC     790
N  Q  I  I  A  N  G  G  N  T  V  K  A  L  G  L  K  K  P  L

TTCCACGCTGCCATCGGCTCCTCCGTCTTCCTCCCCTACCAAGCCAAGTACAACTCCCCC     850
F  H  A  A  I  G  S  S  V  F  L  P  Y  Q  A  K  Y  N  S  P

TTCGCCGAGCTGCTCTACTCCCAACTCGTCTCGGCGACAAACTGCACCAAAGCCGCCTCG     910
F  A  E  L  L  Y  S  Q  L  V  S  A  T  N  C  T  K  A  A  S

TCCTTCGCTTGCCTCGAAGCTGTCGACGCTGCGGCGCTCGCTGCGGCGGGCGTGAAGAAC     970
S  F  A  C  L  E  A  V  D  A  A  A  L  A  A  A  G  V  K  N

TCGGCGGCGTTCCCGTTCGGGTTTTGGTCGTATGTCCCGGTCGTCGACGGGACCTTCTTG    1030
S  A  A  F  P  F  G  F  W  S  Y  V  P  V  V  D  G  T  F  L
```

FIG. 8A

ACTGAGCGCGCGTCGCTCCTTCTCGCCAAGGGCAAGAAGAACCTCAATGGCAACCTCTTC 1090
T E R A S L L L A K G K K N L N G N L F

ACCGGGATCAACAACCTCGACGAAGGATTCATATTCACTGACGCCACTATTCAGAACGAC 1150
T G I N N L D E G F I F T D A T I Q N D

ACGATCAGCGACCAGTCGCAGCGCGTCTCCCAGTTCGACCGCCTCCTCGCCGGCCTCTTC 1210
T I S D Q S Q R V S Q F D R L L A G L F

CCCTACATCACCTCGGAGGAGCGCCAGGCCGTCGCGAAGCAGTACCCGATCTCCGACGCG 1270
P Y I T S E E R Q A V A K Q Y P I S D A

CCGTCAAAGGGCAACACCTTCTCTCGCATCTCGGCCGTCATCGCGGACTCGACCTTCGTC 1330
P S K G N T F S R I S A V I A D S T F V

TGCCCGACCTACTGGACCGCCGAGGCGTTCGGCTCGTCCGCCCACAAGGGCCTCTTCGAC 1390
C P T Y W T A E A F G S S A H K G L F D

TACGCGCCGGCTCACCACGCGACCGACAACTCGTACTACATCGGCTCCATCTGGAACGGC 1450
Y A P A H H A T D N S Y Y I G S I W N G

AAGAAGTCGGTCTCGTCCGTCCAGTCCTTCGACGGCGCGCTCGGCGGCTTCATCGAGACG 1510
K K S V S S V Q S F D G A L G G F I E T

TTCAACCCGAACAACAACGCTGCCAACAAGACCATCAACCCTTACTGGCCGACGTTCGAC 1570
F N P N N N A A N K T I N P Y W P T F D

TCGGGCAAGCAGCTCCTCTTCAACACGACGACGAGGGACACCCTCTCTCCCGCCGACCCG 1630
S G K Q L L F N T T T R D T L S P A D P

CGCATCGTTGAGACTTCAAGCTTGACCGACTTTGGCACGAGCCAGAAGACCAAGTGCGAC 1690
R I V E T S S L T D F G T S Q K T K C D

TTCTGGCGTGGGTCAATCTCGGTGAACGCGGGTCTC 1726
F W R G S I S V N A G L

FIG. 8B

```
GGATCCACCCGAACTCTGTCCCGCTTTCTGGCTTTCTTCCTTGCTGTCGCCCCATCGCCT      60

                |-- Translation Start -->
TTCCCGACTCGCCGCCATGCTCCTTAACCTCTTCACCCTCGCCTCCCTCGCTGCGACGCT     120
                M  L  L  N  L  F  T  L  A  S  L  A  A  T  L

                                                |- Mature peptide ->
CCAGCTCGCCTTTGCCTCTCCGACCTCCCTCGTCCGCCGCACGAACCCAAACGAGCCCCC     180
 Q  L  A  F  A  S  P  T  S  L  V  R  R  T  N  P  N  E  P  P

TCCCGTCGTCGACCTCGGCTACGCCCGCTACCAAGGCTACTTGAACGAGACCGCCGGACT     240
 P  V  V  D  L  G  Y  A  R  Y  Q  G  Y  L  N  E  T  A  G  L

CTACTGGTGGCGCGGAATCCGCTACGCCTCGGCTCAGCGCTTCCAGGCTCCTCAGACGCC     300
 Y  W  W  R  G  I  R  Y  A  S  A  Q  R  F  Q  A  P  Q  T  P

CGCGACGCACAAGGCCGTCCGCAACGCGACTGAGTATGGACCGATCTGTTGGCCGGCTAG     360
 A  T  H  K  A  V  R  N  A  T  E  Y  G  P  I  C  W  P  A  S

CGAGGGAACCAACACGACCAAGGGCTTGCCGCCGCCTAGCAACAGCTCGAGCAGCGCGCC     420
 E  G  T  N  T  T  K  G  L  P  P  P  S  N  S  S  S  S  A  P

GCAGAAACAGGCGTCGGAGGATTGCCTCTTCCTCAATGTCGTTGCCCCCGCCGGCTCGTG     480
 Q  K  Q  A  S  E  D  C  L  F  L  N  V  V  A  P  A  G  S  C

CGAGGGCGACAATCTTCCCGTCCTCGTCTACATTCACGGAGGTGGCTACGCCTTCGGCGA     540
 E  G  D  N  L  P  V  L  V  Y  I  H  G  G  G  Y  A  F  G  D

TGCGAGCACCGGCAGCGACTTTGCCGCCTTCACCAAGCACACGGGAACCAAGATGGTCGT     600
 A  S  T  G  S  D  F  A  A  F  T  K  H  T  G  T  K  M  V  V

TGTAAATCTCCAGTACCGTCTCGGCAGCTTTGGTTTCCTCGCTGGCCAAGCCATGAAGGA     660
 V  N  L  Q  Y  R  L  G  S  F  G  F  L  A  G  Q  A  M  K  D

                                        [---- Intron #1 ----------
CTACGGTGTAACGAACGCCGGCTTGCTTGACCAGGTGAGTTTCCCGCATGATACCCGCCC     720
 Y  G  V  T  N  A  G  L  L  D  Q

-------------------------------------]
ACCTTTCGACTCATGCTGACGCCTCTCCCGCTCGCAGCAATTCGCCCTTCAATGGGTTCA     780
                                        Q  F  A  L  Q  W  V  Q

ACAGCACGTCTCGAAGTTCGGCGGCAACCCCGATCACGTTACGATTTGGGGCGAGTCTGC     840
 Q  H  V  S  K  F  G  G  N  P  D  H  V  T  I  W  G  E  S  A

                                        [---- Intron #2 -------
AGGCGCAGGGTCCGTTATGAACCAGATCATTGCGAACGTGAGCCACCCGAACCGATCTCC     900
 G  A  G  S  V  M  N  Q  I  I  A  N

-------------------------------------------------]
AGCCGACTTTCCCCCCCCCCCCCCCCCCGCTGACCTCCCTCGTCTTGCAGGGCGGCAACA     960
                                                    G  G  N  T

CCGTCAAGGCTCTCGGTCTCAAGAAGCCCCTCTTCCACGCTGCCATCGGCTCCTCCGTCT    1020
  V  K  A  L  G  L  K  K  P  L  F  H  A  A  I  G  S  S  V  F

TCCTCCCCTACCAAGCCAAGTACAACTCCCCCTTCGCCGAGCTGCTCTACTCCCAACTCG    1080
  L  P  Y  Q  A  K  Y  N  S  P  F  A  E  L  L  Y  S  Q  L  V
```

FIG. 9A

```
CTGCGGCGCTCGCTGCGGCGGGCGTGAAGAACTCGGCGGCGTTCCCGTTCGGGTTTTGGT   1200
  A  A  L  A  A  A  G  V  K  N  S  A  A  F  P  F  G  F  W  S

CGTATGTCCCGGTCGTCGACGGGACCTTCTTGACTGAGCGCGCGTCGCTCCTTCTCGCCA   1260
  Y  V  P  V  V  D  G  T  F  L  T  E  R  A  S  L  L  L  A  K

                          [---- Intron #3 --------------------
AGGGCAAGAAGAACCTCAATGGCGTGCGTGGCGAGCTTTCGAGTGCTTCAGGATCTCGCT   1320
  G  K  K  N  L  N  G

----------------------]                                    [---
GACACTGTCGACCGGCTCGCAGAACCTCTTCACCGGGATCAACAACCTCGACGAAGATGA   1380
                       N  L  F  T  G  I  N  N  L  D  E  G

---- Intron #4 ----------------------------------------------]
GTTCCCGTCGACGGCTCTGTTCGCCCAGCGAGACTGACTTGTTCTTTTGCGAAGATTACG   1440

ATTCATATTCACTGACGCCACTATTCAGAACGACACGATCAGCGACCAGTCGCAGCGCGT   1500
  F  I  F  T  D  A  T  I  Q  N  D  T  I  S  D  Q  S  Q  R  V

CTCCCAGTTCGACCGCCTCCTCGCCGGCCTCTTCCCCTACATCACCTCGGAGGAGCGCCA   1560
  S  Q  F  D  R  L  L  A  G  L  F  P  Y  I  T  S  E  E  R  Q

GGCCGTCGCGAAGCAGTACCCGATCTCCGACGCGCCGTCAAAGGGCAACACCTTCTCTCG   1620
  A  V  A  K  Q  Y  P  I  S  D  A  P  S  K  G  N  T  F  S  R

                                    [---- Intron #5 -----------
CATCTCGGCCGTCATCGCGGACTCGACCTTCGTGTGCGTTCCCCGTCGTCTTCTCCGAGT   1680
  I  S  A  V  I  A  D  S  T  F  V

----------------------------]
ATTCCGCTGACTTCCCGCTTGCCCGCAGCTGCCCGACCTACTGGACCGCCGAGGCGTTCG   1740
                             C  P  T  Y  W  T  A  E  A  F  G

GCTCGTCCGCCCACAAGGGCCTCTTCGACTACGCGCCGGCTCACCACGCGACCGACAACT   1800
  S  S  A  H  K  G  L  F  D  Y  A  P  A  H  H  A  T  D  N  S

CGTACTACATCGGCTCCATCTGGAACGGCAAGAAGTCGGTCTCGTCCGTCCAGTCCTTCG   1860
  Y  Y  I  G  S  I  W  N  G  K  K  S  V  S  S  V  Q  S  F  D

ACGGCGCGCTCGGCGGCTTCATCGAGACGTTCAACCCGAACAACAACGCTGCCAACAAGA   1920
  G  A  L  G  G  F  I  E  T  F  N  P  N  N  N  A  A  N  K  T

CCATCAACCCTTACTGGCCGACGTTCGACTCGGGCAAGCAGCTCCTCTTCAACACGACGA   1980
  I  N  P  Y  W  P  T  F  D  S  G  K  Q  L  L  F  N  T  T  T

CGAGGGACACCCTCTCTCCCGCCGACCCGCGCATCGTTGAGACTTCAAGCTTGACCGACT   2040
  R  D  T  L  S  P  A  D  P  R  I  V  E  T  S  S  L  T  D  F

TTGGCACGAGCCAGAAGACCAAGTGCGACTTCTGGCGTGGGTCAATCTCGGTGAACGCGG   2100
  G  T  S  Q  K  T  K  C  D  F  W  R  G  S  I  S  V  N  A  G

GTCTCTAGGCGTCTTTCCTTCCGACTTCCTTCGTTCTTTCGTTGTTTATTCTTGCAGTTC   2160
  L  *

CGTTGTATCGGCCATTCGTGCGTGTAGCTCACTCGAGTATAGACGTTGGCAAGTGCGAAA   2220
```

FIG. 9B

TRN 2-1738 RHODOSPORIDIUM ESTERASE cDNA$

⌜Translation Start ⌜Mature Peptide

MLLNLFTLASLAATLQLAFASPTSLVRRTNPNEPPPVVDLGYARYQGYLNETAGLYWWRG

IRYASAQRFQAPQTPATHKAVRNATEYGPICWPASEGTNTTKGLPPPSNSSSSAPQKQAS

EDCLFLNVVAPAGSCEGDNLPVLVYIHGGGYAFGDASTGSDFAAFTKHTGTKMVVVNLQY

RLGSFGFLAGQAMKDYGVTNAGLLDQQFALQWVQQHVSKFGGNPDHVTIWGESAGAGSVM

NQIIANGGNTVKALGLKKPLFHAAIGSSVFLPYQAKYNSPFAELLYSQLVSATNCTKAAS

SFACLEAVDAAALAAAGVKNSAAFPFGFWSYVPVVDGTFLTERASLLLAKGKKNLNGNLF

TGINNLDEGFIFTDATIQNDTISDQSQRVSQFDRLLAGLFPYITSEERQAVAKQYPISDA

PSKGNTFSRISAVIADSTFVCPTYWTAEAFGSSAHKGLFDYAPAHHATDNSYYIGSIWNG

KKSVSSVQSFDGALGGFIETFNPNNNAANKTINPYWPTFDSGKQLLFNTTTRDTLSPADP

RIVETSSLTDFGTSQKTKCDFWRGSISVNAGL*

FIG. 10

Amino acid composition from 1 to 572
TRN 2-1738 RHODOSPORIDIUM ESTERASE cDNA

	Total	Percent
A	67	11.7
C	7	1.2
D	25	4.4
E	16	2.8
F	35	6.1
G	49	8.6
H	9	1.6
I	21	3.7
K	25	4.4
L	48	8.4
M	4	0.7
N	35	6.1
P	31	5.4
Q	26	4.5
R	16	2.8
S	52	9.1
T	43	7.5
V	32	5.6
W	10	1.7
Y	21	3.7
Acidic	41	7.2
Basic	41	7.2
Charged	82	14.3
Net charge	0	0.0
Hydrophobic	136	23.8
Residues	572	
MW	61334	

FIG. 11